FORM PTO-1390 (Modified) (REV 11-2000) U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER	
217941US0PCT	
U.S. APPLICATION NO. (IF KNOWN, SEE 37 CFR)	10/030101

INTERNATIONAL APPLICATION NO	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/EP00/06916	20 July 2000	23 July 1999

TITLE OF INVENTION
FORMULATIONS OF STEROID SOLUTIONS FOR INHALATORY ADMINISTRATION

APPLICANT(S) FOR DO/EO/US
MALVOLTI Chiara et al.

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☒ This is an express request to begin national examination procedures (35 U.S.C. 371(f)) The submission must include items (5), (6), (9) and (24) indicated below.
4. ☒ The US has been elected by the expiration of 19 months from the priority date (Article 31).
5. ☒ A copy of the International Application as filed (35 U.S.C. 371 (c) (2))
   a. ☐ is attached hereto (required only if not communicated by the International Bureau).
   b. ☒ has been communicated by the International Bureau.
   c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☐ An English language translation of the International Application as filed (35 U S.C. 371(c)(2)).
   a. ☐ is attached hereto.
   b. ☐ has been previously submitted under 35 U.S.C. 154(d)(4).
7. ☒ Amendments to the claims of the International Application under PCT Article 19 (35 U S.C. 371 (c)(3))
   a. ☐ are attached hereto (required only if not communicated by the International Bureau).
   b. ☐ have been communicated by the International Bureau.
   c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   d. ☒ have not been made and will not be made.
8. ☐ An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ☐ An oath or declaration of the inventor(s) (35 U.S.C. 371 (c)(4)).
10. ☐ An English language translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371 (c)(5)).
11. ☒ A copy of the International Preliminary Examination Report (PCT/IPEA/409)
12. ☒ A copy of the International Search Report (PCT/ISA/210).

Items 13 to 20 below concern document(s) or information included:

13. ☒ An Information Disclosure Statement under 37 CFR 1 97 and 1.98.
14. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
15. ☒ A **FIRST** preliminary amendment.
16. ☐ A **SECOND** or **SUBSEQUENT** preliminary amendment.
17. ☐ A substitute specification.
18. ☐ A change of power of attorney and/or address letter.
19. ☐ A computer-readable form of the sequence listing in accordance with PCT Rule 13ter.2 and 35 U.S.C. 1.821 - 1.825.
20. ☐ A second copy of the published international application under 35 U.S.C. 154(d)(4).
21. ☐ A second copy of the English language translation of the international application under 35 U.S.C. 154(d)(4).
22. ☐ Certificate of Mailing by Express Mail
23. ☒ Other items or information:

Notice of Priority/Form PTO-1449

U.S. APPLICATION NO (IF KNOWN, SEE 37 CFR	INTERNATIONAL APPLICATION NO	ATTORNEY'S DOCKET NUMBER
10/030101	PCT/EP00/06916	217941US0PCT

24. The following fees are submitted:.	CALCULATIONS PTO USE ONLY
BASIC NATIONAL FEE (37 CFR 1.492 (a) (1) - (5)) :	
☐ Neither international preliminary examination fee (37 CFR 1 482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO **$1040.00**	
☒ International preliminary examination fee (37 CFR 1.482) not paid to USPTO but International Search Report prepared by the EPO or JPO **$890.00**	
☐ International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1 445(a)(2)) paid to USPTO **$740.00**	
☐ International preliminary examination fee (37 CFR 1.482) paid to USPTO but all claims did not satisfy provisions of PCT Article 33(1)-(4) **$710.00**	
☐ International preliminary examination fee (37 CFR 1.482) paid to USPTO and all claims satisfied provisions of PCT Article 33(1)-(4) **$100.00**	
ENTER APPROPRIATE BASIC FEE AMOUNT =	**$890.00**
Surcharge of **$130.00** for furnishing the oath or declaration later than ☐ 20 ☒ 30 months from the earliest claimed priority date (37 CFR 1.492 (e)).	**$130.00**

CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE		
Total claims	11 - 20 =	0	x $18 00	**$0.00**	
Independent claims	1 - 3 =	0	x $84.00	**$0.00**	
Multiple Dependent Claims **(check if applicable)**.			☐	**$0.00**	
TOTAL OF ABOVE CALCULATIONS =				**$1,020.00**	
☐ Applicant claims small entity status. See 37 CFR 1.27). The fees indicated above are reduced by 1/2.				**$0.00**	
SUBTOTAL =				**$1,020.00**	
Processing fee of **$130.00** for furnishing the English translation later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492 (f)). +				**$0.00**	
TOTAL NATIONAL FEE =				**$1,020.00**	
Fee for recording the enclosed assignment (37 CFR 1.21(h)) The assignment must be accompanied by an appropriate cover sheet (37 CFR 3 28, 3.31) **(check if applicable).** ☐				**$0.00**	
TOTAL FEES ENCLOSED =				**$1,020.00**	
				Amount to be: refunded	$
				charged	$

a. ☒ A check in the amount of **$1,020.00** to cover the above fees is enclosed.

b. ☐ Please charge my Deposit Account No. _____ in the amount of _____ to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. **15-0030** A duplicate copy of this sheet is enclosed.

d. ☐ Fees are to be charged to a credit card. **WARNING:** Information on this form may become public. **Credit card information should not be included on this form.** Provide credit card information and authorization on PTO-2038.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

Surinder Sachar
Registration No. 34,423

22850

Surinder Sachar
SIGNATURE

Norman F. Oblon
NAME

24,618
REGISTRATION NUMBER

Jan 17 2002
DATE

Docket No. 217941US0PCT
IN RE APPLICATION OF: Chiara MALVOLTI et al.
SERIAL NO: New U.S. PCT Application (Based on PCT/EP00/06916
FILED: HEREWITH
FOR: FORMULATIONS OF STEROID SOLUTIONS FOR INHALATORY ADMINISTRATION

ASSISTANT COMMISSIONER FOR PATENTS
WASHINGTON, D.C. 20231

SIR:

Transmitted herewith is an amendment in the above-identified application.

☒ No additional fee is required

☐ Small entity status of this application under 37 C.F.R. §1.9 and §1.27 is claimed.

☒ Additional documents filed herewith: PCT Transmittal Letter/Notice of Priority/International Preliminary Examination Report/International Search Report/Form PTO-1449/Information Disclosure Statement/Check for $1,020.00

The Fee has been calculated as shown below:

CLAIMS	CLAIMS REMAINING		HIGHEST NUMBER PREVIOUSLY PAID	NO. EXTRA CLAIMS	RATE	CALCULATIONS
TOTAL	11	MINUS	20	0	× $18 =	$0.00
INDEPENDENT	1	MINUS	3	0	× $84 =	$0.00
		☐ MULTIPLE DEPENDENT CLAIMS			+ $280 =	$0.00
		TOTAL OF ABOVE CALCULATIONS				$0.00
		☐ Reduction by 50% for filing by Small Entity				$0.00
		☐ Recordation of Assignment			+ $40 =	$0.00
					TOTAL	$0.00

☐ A check in the amount of **$0.00** is attached.

☒ Please charge any additional Fees for the papers being filed herewith and for which no check is enclosed herewith, or credit any overpayment to deposit Account No. 15-0030. A duplicate copy of this sheet is enclosed.

☒ If these papers are not considered timely filed by the Patent and Trademark Office, then a petition is hereby made under 37 C.F.R. §1.136, and any additional fees required under 37 C.F.R. §1.136 for any necessary extension of time may be charged to Deposit Account No. 15-0030. A duplicate copy of this sheet is enclosed.

OBLON, SPIVAK, McCLELLAND,
MAIER & NEUSTADT, P.C.

Norman F. Oblon
Registration No. 24,618

Surinder Sachar
Registration No. 34,423

22850
Customer Number 22850
Tel. (703) 413-3000
Fax. (703) 413-2220
(OSMMN 10/01)

#4/a

217941US-0PCT

IN THE UNITED STATES PATENT & TRADEMARK OFFICE

IN RE APPLICATION OF :

CHIARA MALVOLTI ET AL : ATTN: APPLICATION DIVISION

SERIAL NO: NEW U.S. PCT APPLN :
(Based on PCT/EP00/06916)

FILED: HEREWITH :

FOR: FORMULATIONS OF STEROID :
SOLUTIONS FOR INHALATORY
ADMINISTRATION

PRELIMINARY AMENDMENT

ASSISTANT COMMISSIONER FOR PATENTS
WASHINGTON, D.C. 20231

SIR:

Prior to examination on the merits, please amend the above-identified application as follows.

IN THE CLAIMS

Please amend the claims as shown in the marked-up copy following this amendment to read as follows.

2. (Amended) The formulation according to claim 1, wherein the carrier consists of water and propylene glycol in a 50:50 v/v ratio.

3. (Amended) The formulation according to claim 1, wherein pH ranges from 4.0 to 4.5 and has been corrected by using HC1.

4. (Amended) The formulation according to claim 1, wherein the steroid is an acetal derivative or an acetonide derivative.

5. (Amended) The formulation according to claim 1, wherein the acetal derivative is budesonide or the epimers thereof.

6. (Amended) The formulation according to claim 1, wherein the acetonide derivative is flunisolide.

7. (Amended) The formulation according to claim 5, wherein budesonide concentration ranges from 0.025 to 0.05%.

8. (Amended) The formulation according to claim 6, wherein flunisolide concentration is 0.1%.

9. (Amended) The formulation according to claim 1, wherein the osmolarity is not more than 7500 mOsm/1.

10. (Amended) The formulation according to claim 1, stable according to the requirements of the Guidelines for medicinal products for human use.

REMARKS

Claims 1-11 are active in the present application. Claims 2-10 have been amended to remove multiple dependencies and for clarity. No new matter is added. An action on the merits and allowance of claims is solicited.

Respectfully submitted,

OBLON, SPIVAK, McCLELLAND,
MAIER & NEUSTADT, P.C.

Norman F. Oblon
Attorney of Record
Registration No. 24,618

Stefan U. Koschmieder, Ph.D.
Registration No. 50,238

Surinder Sachar
Registration No. 34,423

22850

(703) 413-3000
Fax #: (703)413-2220
NFO:SUK\la
I:\atty\SUKOS\217941us-pr.wpd

217941US-0PCT

Marked-Up Copy
Serial No:

Amendment Filed on:
1-17-02

IN THE CLAIMS

Please amend the claims as follows.

--2. (Amended) [A] The formulation according to claim 1, wherein the carrier consists of water and propylene glycol in a 50:50 v/v ratio.

3. (Amended) [A] The formulation according to [claims 1 and 2] claim 1, wherein pH ranges from 4.0 to 4.5 and has been corrected by using HC1.

4. (Amended) [A] The formulation according to [claims 1-3] claim 1, wherein the steroid is an acetal derivative or an acetonide derivative.

5. (Amended) [A] The formulation according to [claims 1-4] claim 1, wherein the acetal derivative is budesonide or the epimers thereof.

6. (Amended) [A] The formulation according to [claims 1-4] claim 1, wherein the acetonide derivative is flunisolide.

7. (Amended) [A] The formulation according to claim 5, wherein budesonide concentration ranges from 0.025 to 0.05%.

8. (Amended) [A] The formulation according to claim 6, wherein flunisolide concentration is 0.1%.

9. (Amended) [A] The formulation according to [any preceding claim] claim 1 wherein the osmolarity is not more than 7500 mOsm/1.

10. (Amended) [A] The formulation according to [any preceding claim] claim 1, stable according to the requirements of the Guidelines for medicinal products for human use.--

FORMULATIONS OF STEROID SOLUTIONS FOR INHALATORY ADMINISTRATION

The present invention relates to optimized formulations for nebulisation administration containing antiinflammatory glucocorticoids in hydroalcoholic solution and a process for the preparation thereof.

More particularly, the invention relates to formulations for monodose or multidose vials in the form of preservative-free stable solutions, well-tolerated by the patients, of reduced osmolarity and that can effectively be nebulised with the nebulisers currently available on the market.

PRIOR ART

The administration of drugs through nebulisation has been used for many years and is the mainstay of treatment of diseases which hamper breathing, such as asthma and chronic bronchitis.

One of the advantages of the inhalatory route over the systemic one is the possibility of delivering the drug directly at site of action, avoiding any systemic side-effects, thus resulting in a more rapid clinical response and a higher therapeutic index.

Among the various drugs active on the respiratory system, corticosteroids such as beclomethasone dipropionate, fluticasone propionate, flunisolide and budesonide are of great importance. Said drugs may be administered in the form of pressurized aerosols or by using ultrasonic or jet

nebulisers.

As far as the administration by jet nebulisers is concerned, usually the steroid is either suspended in micronised form in saline or dissolved in water-alcoholic mixtures in the presence of excipients such as buffering agents, stabilizing agents and preservatives.

In particular, budesonide, one of the steroids most applied by means of this administration route by virtue of its better topical/systemic activity ratio, is commercially available only as an aqueous suspension (Pulmicort®), further containing citric acid, sodium citrate, polysorbate 80 and sodium edetate.

In general, suspensions are intrinsically less homogeneous than solutions; furthermore, problems of physical stability can arise during storage, due to the formation of agglomerates or cakes which are difficult to be redispersed.

Said drawback can in turn give rise to problems of repartition and so of dosage uniformity during the filling of the containers; beside that, the lack of homogeneity could also compromise the correct posology of the drug or at least cause a therapeutically less effective administration, since the transfer of the dose from the container to the nebuliser reservoir by the patient could be incomplete.

Furthermore, the effectiveness of the administration form depends on the deposition of an adequate amount of particles at the site of action. One of the most critical

parameters determining the proportion of inhalable drug which will reach the lower respiratory tract of a patient is the size of the particles emerging from the device. In order to ensure an effective penetration into the bronchioli and alveoli and hence ensure a high respirable fraction, the mean aerodynamic diameter (MAD) of the particles should be lower than 6 microns (μm).

Particles with higher MAD are in fact deposited in the higher respiratory tract, i.e. the oropharynx and may give rise to topical side effects; otherwise they may be absorbed thus giving rise to systemic side effects.

In this respect, it is difficult for aqueous suspensions to maintain a constant particle size distribution during their shelf life; in the prior art (Davis S *et al* Int J Pharm 1, 303-314, 1978; Tiano S *et al* Pharm Dev Tech 1, 261-268, 1996; Taylor K *et al* Int J Pharm 153, 93-104, 1997) it is indeed reported that as environmental humidity conditions change, the suspended particles can grow in size following partial or complete recrystallization of the even small amount of solute dissolved, therefore increasing in MAD; said increase may, in turn, impair both the nebulisation efficiency, which is inversely proportional to the MAD of the particles, and the therapeutical efficacy, as particles with MAD greater than 6 μm cannot be delivered to the preferential site of action.

Steroids such as beclomethasone or fluticasone can only be acceptably formulated as a suspension.

Other glucocorticosteroids such as budesonide or

flunisolide can be also prepared as a solution, but, due to their high lipophilicity, it is not possible to prepare simple solutions having the desired concentration of active ingredient without using a suitable co-solvent such as propylene glycol, glycerol or polyethylene glycol. Said co-solvents are however less volatile than water; consequently, by increasing the osmolarity they decrease the surface tension of the whole solution so slowing down the evaporation rate of the droplets produced by nebulisation. This gives rise to a high percentage of particles of size greater than 6 μm.

In the solution formulations currently available on the market such as those containing flunisolide, the carrier is usually a mixture of physiological solution (0.9% saline in water) and propylene glycol. The presence of sodium chloride contributes to significantly increase the osmolarity and the ionic strength of the solution which may result in an even higher percentage of non respirable particles , being the formulations not effectively aerosolized by the common nebulizers. An excessive hypertonicity can also induce tolerability problems in the patient, which are paradoxically manifested by cough and bronchospasm (O'Callaghan C *et al* Lancet, ii, 1424-1425,1986).

Inhalatory formulations should meet a further important requirement, which is a pharmaceutically acceptable shelf-life. In order to maintain potency, minimize the formation of degradation products and prevent any microbiological contaminations, preservatives and stabilizing agents such as

antioxidants and metal chelating agents are frequently used. The prior art reports that some substances commonly used for this purpose can either induce allergic reactions or give rise to irritation of the respiratory mucosas (Menendez R *et al* J Allergy Clin Immunol 84, 272-274, 1989; Afferty P *et al* Thorax 43, 446-450, 1988).

Moreover, they further increase the osmolarity.

In view of the potential problems and disadvantages connected with the formulations containing anti-inflammatory glucocorticoids currently available on the market, it would be highly advantageous to provide formulations in solution, containing no stabilizing agents and/or preservatives, provided of adequate shelf life, whose osmolarity permits generation of an effective aerosol well tolerated by patients.

DISCLOSURE OF THE INVENTION

The main object of the present invention is to provide solution formulations containing therapeutically effective concentrations of antiinflammatory glucocorticoids, provided of adequate shelf life, without stabilizing agents and preservatives, well tolerated by patients, which can be effectively aerosolized with the common nebulizers and able to ensure a high respirable fraction by producing active ingredient particles with MAD predominantly ranging from 1 to 6 μm.

More specifically, the present invention aims to provide optimized solutions of budesonide, to be administered through nebulisation, without using

preservatives and/or stabilizing agents.

Said aim has been attained by preparing a pharmaceutical formulation, suitable for inhalation through nebulisation, which consists of a solution of a steroid in that:

a) the steroid concentration ranges from 0.01% to 0.1%;

b) the carrier is a mixture of water and propylene glycol in a ratio ranging from 60:40 to 30:70 v/v;

c) pH ranges from 3.5 to 5.0 and has been adjusted by using a concentrated strong acid;

wherein the osmolarity is not more than 7500 mOsm/l, preferably not more than 7000, even more preferably not more than 6800 and the percentage of nebulised active ingredient particles with MAD below 6 μm is higher than 70% and the nebulisation efficiency after 5 minutes is higher than 20%.

In a particular embodiment of the invention, the formulations are prepared by using a carrier consisting of a water : propylene glycol 50:50 v/v mixture, correcting pH with concentrated strong acids such as hydrochloric acid to values preferably ranging from 4 to 5. It has in fact surprisingly found that if pH, instead of being just corrected, is adjusted by addition of the usual saline buffers such as the dibasic sodium phosphate/citric acid couple, the solutions do not remain stable for a pharmaceutically acceptable time. After addition of said buffers, under accelerated stability conditions (40°C, 75% relative humidity [R.H.]), a 10% or higher loss of the assay

is in fact observed already after three months. Conversely, the assay of the active ingredient in the solutions whose pH has been simply corrected to 4.0 or 4.5 with HCl remains substantially unchanged after 18 months under long term conditions (25°C, 60° R.H.) and only a slight decrease in the assay is observed after 6 months under accelerated conditions. The solutions of the invention require no addition of stabilizing agents such as metal chelating agents or other antioxidants.

Although it is known from the prior art that the stability of steroids bearing a dihydroxyacetone side chain, such as budesonide and flunisolide, depends on pH and that said steroids are more stable within a pH range of 3-5 (Das Gupta V, J Pharm Sci 12, 1453, 1983; Timmins P *et al.* J Pharm Pharmacol 35, 175, 1983), stable budesonide in solution in a simple water-alcoholic mixture consisting of water and propylene glycol has never been reported; moreover it has never been disclosed that stability depends so dramatically on the way of adjusting the pH.

Analogously it has never been reported that said solutions can be efficaciously delivered by means of a nebulizer to the lower respiratory tract.

The pH of the formulation also affects the tolerability of the nebulised solution. Aerosol formulations with pH ranging from 4 to 5 are recognizedly well tolerated by the patient (Morén F *et al*, Aerosol in Medicine, Elsevier, Amsterdam, 1993, page 342). Furthermore, the simple correction of pH with strong acids causes a decrease in the

buffering properties of the solution thereby allowing the pH of the droplets to readily change and attain more physiologically acceptable values once the pulmonary area has been reached. On the other hand, the correction of the natural pH of the water to lower values is extremely advantageous when the solution is stored in glass ampoules as the pH inside such containers tends to increase during storage thus adversely affecting the stability of the active ingredient.

The only known solution formulation of budesonide commercially available is a lotion for topical use containing almost 80% w/w of alcohols.

This kind of formulation, due to the so high content of alcohols, is clearly not suitable for inhalatory purposes.

EP 794767 (Falk) discloses budesonide solutions at pH below 6 to be used in the preparation of enemas and rectal foams. The formulations are claimed as stable, but actually if we look at the examples only water-alcoholic formulations involving the use of antioxidants such as sodium edetate or complexing agents such as cyclodextrins are stable for a pharmaceutically acceptable time (at least 6 months).

When such preservatives are not present, the assay of the active ingredient decreases by more than 30% already after 4 weeks at 40°C. The minimum stability requirements prescribed by the Guidelines for medicinal products for human use - Quality and biotechnology, Vol 3A, 1998 Edition, pages 127-134, envision a loss of assay of the active ingredient lower than 5% after storage under accelerated

conditions (40°C, 75% R.H.) for six months. In EP 794767, simple 0.0033% solutions in water at different pH values, have been tested only after 14 day of storage . Solutions in propylene glycol alone, which is anyway a carrier unsuitable for the administration *via* nebulisation, are found to be sufficiently stable only at pH 2.8. Therefore EP 794767 does not teach to prepare pharmaceutically acceptable hydro-alcoholic budesonide solution formulation stable without the aid of stabilizing agents which may be efficiently nebulized.

DE 19625027 claims solutions of drugs such as flunisolide and budesonide, stable by addition of an organic or inorganic acid, for the preparation of pressurized aerosols, using as a propellant a carrier containing at least 70% of ethanol. Said solutions, due to the high percentage of ethanol, which is recognisedly irritant, are not suitable for nebulisation and always contain EDTA.

In the solutions of the invention, consisting of a physiologically acceptable selected range of propylene glycol in a ratio to water ranging from 60:40 to 30:70 v/v it is also possible to avoid the use of preservatives, as it is proved by the bioburden which remains within the limits provided for by the European Pharmacopoeia during the whole stability time of the product.

Since the solutions of the invention are stable without the use of stabilizing agents and preservatives, it is possible to keep their osmolarity to a lower value with respect to known solution formulations, in such a way as to

give rise to either an improved efficiency of nebulization and an increased fraction of respirable droplets.

It has in fact been found, and this is a further object of the invention, that the formulations consisting of simple water : propylene glycol solutions are more efficiently nebulised than the corresponding solutions containing sodium chloride and/or salts acting as buffering or stabilizing agents. Furthermore, the formulations of the invention can deliver a higher amount of active ingredient with MAD ranging from 1 to 6 μm therefore providing a larger respirable fraction.

Davis S in Int. J. Pharm. 1, 71-83, 1978 reports that when a water : propylene glycol mixture is used for nebulising 0.1% of flunisolide, the optimal percentage of glycol to attain efficient nebulisation is around 50-60% v/v, but no teachings as regards the preparation of stable solutions in said carrier without further addition of stabilizing agents or buffering salt is reported. Furthermore, in Int. J. Pharm. 1, 85-83, 1978, in a study aimed at evaluating as a carrier the water-propylene glycol-ethanol system, Davis suggests that the presence of alcohol would increase the total output from the nebuliser. As it can be appreciated from Table 2 of the same paper, the nebulisation efficiency of the solution with no alcohol is indeed rather low (1 ml in 21 min).

Derbacher J (Atemwegs-Lungenkrank 20, 381-82, 1994), in a study which emphasizes the importance of pH and osmolarity of solutions for the inhalatory route, reports, inter alia,

a budesonide isotonic solution (282 mosm/l) with pH 4, but no information are given concerning the composition of the carrier. Moreover it is not reported whether either the concentration or stability of the active ingredient are suitable for a pharmaceutical use. It is in any case unlikely that budesonide dissolves in an aqueous medium at a therapeutic concentration, due to its high lipophilicity.

With respect to the prior art, the compositions of the invention are therefore characterized by the following features:

- a steroid, preferably consisting of budesonide in solution at concentrations ranging from 0.001% to 0.1%, preferably from 0.025% to 0.05%;
- a carrier consisting of a water : propylene glycol mixture in ratios ranging from 60:40 v/v to 30:70 v/v, preferably 50:50 v/v;
- a pH ranging from 3.5 to 5.0, preferably from 4.0 to 4.5, characterized by a shelf life of at least two years and a reduced osmolarity in such a way as to improve the efficiency of nebulization and the fraction of respirable droplets.

Advantageously the osmolarity is not more than 7500 mOsm/l, preferably not more than 7000, even more preferably not more than 6800, based on the calculation of the depression of the freezing point.

Similar compositions can be prepared with acetonide glucocorticoids and in particular with flunisolide.

Preferred carriers for the formulations of the

invention are those consisting of a water : propylene glycol mixture in ratios ranging from 60:40 to 30:70 v/v, preferably in a 50:50 v/v ratio, the concentration of the active ingredient in the solution ranging from 0.001 to 0.1% by weight.

The pH can be corrected by using any concentrated strong acid such as HCl and should range from 3.5 to 5.0, preferably from 4.0 to 4.5. Preferred active ingredients are steroids usually administered in the inhalatory treatment of respiratory diseases. Particularly preferred are acetonide derivatives such as flunisolide. Even more preferred are acetal derivatives such as budesonide or the epimers thereof.

The obtained solutions can be distributed in suitable containers such as multidose vials for nebulisation or preferably in monodose vials, preformed or produced with a technology capable of guaranteeing filling the vials under inert atmosphere. The solution formulations can be advantageously sterilized by filtration.

The formulations of the invention are illustrated in detail by the following examples.

Example 1

Preparation of 0.05% Budesonide solution at pH 4.0 and stability studies

5 litres of propylene glycol was poured into a mixer and heated up to a temperature of 40-50°C. 5 g (0.05%) of budesonide was added, mixing for about 30 min. After cooling to room temperature, an equal volume of depurated water was

added, stirring for a further 15 minutes. pH of the solution was corrected to 4.0 with 0.1 N HCl. The solution was filtered through a 0.65 mm membrane. The solution was distributed in 2 ml polypropylene monodose vials.

Ingredients

Components	Amounts	
	Total preparation amount	Amount per pharmaceutical unit
Budesonide	5 g	1 mg
Propylene glycol	5 l	1 ml
Depurated water q.s. to	10 l	2 ml
0.1 N HCl q.s. to	about pH 4.0	

The stability of the vials was evaluated both under long-term (25°C, 60% R.H.) and accelerated conditions (40°C, 75% R.H.) [R.H. = relative humidity]. Results are reported in Tables 1 and 2, respectively. Assays of budesonide and of its main related substances (degradation products) were determined by HPLC.

Microbiological controls were carried out according to Eur. Ph. III Ed.

The formulation of the invention turns out to be stable for at least 18 months of storage and no increase in the bioburden is observed. The assay is higher than 97% under long-term conditions, whereas is higher than 95% under accelerated conditions. pH remains substantially unchanged under both conditions. None of the other technological parameters undergoes alterations.

TABLE 1 - Solution of example 1 - Stability under long-term conditions (25°C, 60% R.H.)

Analysis	TECHNOLOGICAL CONTROLS		CHEMICAL CONTROLS			
	solution appearance	packaging appearance	Budesonide (g/100 ml)	Assay (%)	Impurities and degraded (% area)	pH
Confidence limits	Clear colourless solution	Colourless monodose	0.0450-0.0525	95-105	-	-
t= 0	Clear colourless solution	Colourless monodose	0.0513	100	0.83	3.92
t= 1 month	Clear colourless solution	Colourless monodose	0.0507	98.8	0.60	3.89
t= 3 months	Clear colourless solution	Monodose colourless	0.0514	100.2	0.82	4.00
t= 6 months	Clear colourless solution	Colourless monodose	0.0507	98.8	1.58	3.91
t= 12 months	Clear colourless solution	Colourless monodose	0.0510	99.4	2.17	3.85
t= 18 months	Clear colourless solution	Colourless monodose	0.0500	97.5	2.47	3.92

TABLE 2 - Solution of example 1 - Stability under accelerated conditions (40°C , 75% R.H.)

Analysis	TECHNOLOGICAL CONTROLS		CHEMICAL CONTROLS			
	Solution appearance	Packaging appearance	Budesonide (g/100 ml)	Assay (%)	Impurities and degraded (% area)	pH
Confidence limits	Clear colourless solution	Colourless monodose	0.0450-0.0525	95-105	-	-
t= 0	Clear colourless solution	Monodose colourless	0.0513	100	0.83	3.92
t= 1 month	Clear colourless solution	Monodose colourless	0.0504	98.2	0.84	3.88
t= 2 months	Clear colourless solution	Monodose colourless	0.0506	98.6	1.55	4.01
t= 3 months	Clear colourless solution	Monodose colourless	0.0501	97.7	2.00	3.97
t= 6 months	Clear colourless solution	Monodose colourless	0.0491	95.7	4.07	3.89

Example 2

Preparation of 0.05% Budesonide solution at pH 4.5 and stability tests

According to the process reported in example 1, a solution having the following formula was prepared:

Components	Total amount of the preparation	Amount per pharmaceutical unit
Budesonide	5 g	1 mg
Propylene glycol	5 l	1 ml
Depurated water q.s. to	10 l	2 ml
0.1 N HCl q.s. to about pH 4.5		-

The stability of the monodose vials was evaluated both under long-term (25°C, 60% R.H.) and accelerated conditions (40°C, 75% R.H.).

The results are reported in Tables 3 and 4, respectively.

The determination of the parameters was carried out as reported in example 1.

The formulation of the invention turns out to be stable for at least 18 months of storage and no increase in the bioburden is observed. Under long-term conditions the assay is higher than 97%, whereas under accelerated conditions was higher than 96%. pH remains substantially unchanged under both conditions. None of the other technological parameters

undergoes alterations.

TABLE 3 - Solution of example 2 - Stability under long-term (25°C , 60% R.H.)

Analysis	TECHNOLOGICAL CONTROLS		CHEMICAL CONTROLS			
	Solution appearance	packaging appearance	Budesonide (g/100 ml)	Assay %)	Impurities and degraded (% area)	pH
Confidence limits	Clear colourless solution	Colourless monodose	0.0450-0.0525	95-105	-	-
t= 0	Clear colourless solution	Colourless monodose	0.0508	100	0.88	4.55
t= 1 month	Clear colourless solution	Colourless monodose	0.0505	99.4	0.47	4.44
t= 3 months	Clear colourless solution	Colourless monodose	0.0500	98.4	0.76	4.49
t= 6 months	Clear colourless solution	Colourless monodose	0.0496	97.6	1.22	4.47
t= 12 months	Clear colourless solution	Colourless monodose	0.0500	98.4	1.93	4.32
t= 18 months	Clear colourless solution	Colourless monodose	0.0510	100.4	2.46	4.34

TABLE 4 - Solution of example 2 - Stability under accelerated conditions (40°C , 75% R.H.)

Analysis	TECHNOLOGICAL CONTROLS		CHEMICAL CONTROLS			
	Solution appearance	packaging appearance	Budesonide (g/100 ml)	Assay (%)	Impurities and degraded (% area)	pH
Confidence limits	Clear colourless solution	Colourless monodose	0.0450-0.0525	95-105	-	-
t= 0	Clear colourless solution	Colourless monodose	0.0508	100	0.88	4.55
t= 1 month	Clear colourless solution	Colourless monodose	0.0502	98.8	0.79	4.42
t= 2 months	Clear colourless solution	Colourless monodose	0.0511	100.6	1.62	4.75
t= 3 months	Clear colourless solution	Colourless monodose	0.0496	97.6	1.95	4.48
t= 6 months	Clear colourless solution	Colourless monodose	0.0497	97.8	3.8	4.44

Example 3

Stability comparisons

With a process similar to that described in examples 1 and 2, 0.05% Budesonide reference solutions were prepared whose pH was adjusted by using buffers consisting of different relative percentages of the dibasic sodium phosphate / citric acid couple. Each solution was distributed in 2 ml polypropylene monodose vials (reference solutions 5 to 8). Furthermore, a 0.05% Budesonide solution in saline : propylene glycol 50:50 v/v was prepared whose natural pH was not corrected.

Part of said solution was placed in 2 ml monodose vials (reference solution 4), whereas the remainder was poured into an amber glass ampoule and tightly sealed (reference solution 3).

The vials containing the various solutions and the glass ampoule were stored at 40°C for 6 months. The Budesonide assay and the pH of said samples were evaluated. Results are reported in Table 5.

From the results obtained with the solutions buffered to different pH values a rather high loss of assay can be appreciated already after three months; said solutions are therefore less stable than those described in examples 1 and 2.

Also the solution at natural pH after 6 month storage in monodose vials was less stable than the solutions described in examples 1 and 2 (see Tables 2 and 4); as far as the same solution is concerned, but stored in an amber

glass ampoule, the assay dramatically decreased with an about 20% loss of potency. In this case pH tends to increase during storage to about 6. The loss in the assay is most likely related with the increase of pH.

Therefore the right starting pH value is demonstrated to be of paramount importance for the stability of these formulations. As far as budesonide solutions are concerned, the starting pH needs to be set at a value between 4.0 and 4.5.

TABLE 5 - Comparison solutions of Example 3

Solution	Time 0		1 month		2 months		3 months		6 months	
	(%)	pH	(%)	pH	(%)	pH	(%)	pH	(%)	pH
Sol. 3 - pH 5.7 (glass)*	100.0	5.7	77.4	6.4	-	-	60.8	6.6	52.5	6.1
Sol. 4 - pH 4.7 (monodose)*	100.0	4.7	98.6	4.7	-	-	96.6	4.8	93.6	4.7
Sol. 5 - pH 5.20 buffer	100.0	5.2	-	5.2	-		89.1	5.3	-	
Sol. 6 - pH 4.26 buffer	100.0	4.3	97.4	4.3	95.0	4.3	93.7	4.4	80.4	4.4
Sol. 7 - pH 4.01 buffer	98.6	4.0	96.8	4.0	94.9	4.0	91.9	4.0	-	-
Sol. 8 - pH 3.36 buffer	99.1	3.3	96.7	3.3	94.8	3.4	90.7	3.4	-	-

* natural pH (neither corrected nor buffered)

Example 4

The nebulisation performances of the solution for inhalation described in example 1 were evaluated by multi-stage liquid impinger (M.S.L.I.) analysis, according to the procedure described in Eur. Ph. III Ed., 1997, using a commercial jet nebuliser (PARI-BOY) for a 5 minute nebulisation time. The M.S.L.I. apparatus consists of a number of glass elements mutually connected to form chambers capable of separating the droplets depending on their aerodynamic size. As follows, particles with different size deposit in the various separation chambers.

It is accordingly possible to determine both the nebulisation efficiency (percentage amount of nebulised active ingredient) and the parameters useful to define the respirable fraction, namely the fine particle fraction (amount and relative % of particles of active ingredient of size below 6.8 mm) and extra fine particle fraction (amount and relative % of particles of active ingredient of size below 3 μm).

Monodose vials of the formulation currently available on the market as an aqueous suspension (Pulmicort®) and monodose vials containing the solution 4 of example 3 (saline : propylene glycol 50:50 *v/v*) were nebulised for comparison. Results are reported in Table 6 as a mean of three determinations.

TABLE 6

	Amount of nebulised a.i., μg	Fine particle fraction μg	(%)	Extrafine particle fraction μg	(%)	Efficiency (%)
Solution of ex. 1	366	317	(86.7)	172	(47.2)	31.7
Solution 4 of ex. 3	259	224	(86.5)	127	(49.0)	27.2
Pulmicort®	103	85	(82.5)	47	(45.6)	14.3

a.i. = active ingredient

The results show a significant improvement in terms of nebulisation efficiency and fine and extrafine fractions delivered for the solution of the invention compared with respect to the commercial formulation. An appreciable improvement of said parameters is also observed with respect to the solution in saline : propylene glycol 50:50 *v/v* (solution 4 of ex. 3).

Example 5

The size profile of the droplets produced by nebulisation of the solutions described in examples 1 and 2 was determined by API Aerosizer analysis, using a commercial jet nebuliser (PARI-BOY).

The particle size distribution profile of solution 4 in saline : propylene glycol 50:50 *v/v* described in example 3 was determined for comparison.

Results are reported in Table 7 as diameter (μm) below which respectively 10%, 50% and 90% of the droplets are included.

TABLE 7

	Median aerodynamic diameter of the droplets [MAD] (μm)		
	10%	50%	90%
Solution of ex. 1	2.26	3.86	5.79
Solution of ex. 2	2.03	3.29	4.79
Solution 4 of ex. 3	3.31	5.21	7.48

The results show a significant shift towards lower values of the size profile of the droplets produced by nebulisation of the solutions of the invention compared with those of the solution in saline : propylene glycol 50:50 v/v (solution 4 of ex. 3).

CLAIMS

1. A stable pharmaceutical formulation for inhalation through nebulisation consisting of a solution of a steroid in which:

a) the steroid concentration ranges from 0.01% to 0.1%;

b) the carrier is a mixture of water and propylene glycol in a ratio ranging from 60:40 to 30:70 v/v;

c) pH ranges from 3.5 to 5.0 and has been adjusted by using a concentrated strong acid;

wherein the percentage of nebulised active ingredient particles with MAD below 6 μm is higher than 70% and the nebulisation efficiency is higher than 20%.

2. A formulation according to claim 1, wherein the carrier consists of water and propylene glycol in a 50:50 v/v ratio.

3. A formulation according to claims 1 and 2, wherein pH ranges from 4.0 to 4.5 and has been corrected by using HCl.

4. A formulation according to claims 1-3, wherein the steroid is an acetal derivative or an acetonide derivative.

5. A formulation according to claims 1-4, wherein the acetal derivative is budesonide or the epimers thereof.

6. A formulation according to claims 1-4, wherein the acetonide derivative is flunisolide.

7. A formulation according to claim 5, wherein budesonide concentration ranges from 0.025 to 0.05%.

8. A formulation according to claim 6, wherein flunisolide concentration is 0.1%.

9. A formulation according to any preceding claim wherein

the osmolarity is not more than 7500 mOsm/l.

10. A formulation according to any preceding claim, stable according to the requirements of the Guidelines for medicinal products for human use .

11. A process for the preparation of pharmaceutical formulations for inhalation through nebulisation according to claim 1 wherein:

a) a solution of the active ingredient in propylene glycol at 40-50°C is prepared;

b) the resulting solution is cooled then diluted with water;

c) pH is corrected with a concentrated strong acid;

d) the solution is filtered and distributed in containers.

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
1 February 2001 (01.02.2001)

PCT

(10) International Publication Number
WO 01/07014 A1

(51) International Patent Classification[7]: A61K 9/08, 9/72, 31/58, A61P 11/06

(21) International Application Number: PCT/EP00/06916

(22) International Filing Date: 20 July 2000 (20.07.2000)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
MI99A001625 23 July 1999 (23.07.1999) IT

(71) Applicant *(for all designated States except US)*: **CHIESI FARMACEUTICI S.P.A.** [IT/IT]; Via Palermo, 26/A, I-43100 Parma (IT).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **MALVOLTI, Chiara** [IT/IT]; Via Palermo, 26/A, I-43100 Parma (IT). **GARZIA, Raffaella** [IT/IT]; Via Palermo, 26/A, I-43100 Parma (IT). **BRAMBILLA, Gaetano** [IT/IT]; Via Palermo, 26/A, I-43100 Parma (IT). **CHIESI, Paolo** [IT/IT]; Via Palermo, 26/A, I-43100 Parma (IT).

(74) Agents: **MINOJA, Fabrizio** et al.; Bianchetti Bracco Minoja S.r.l., Via Rossini, 8, I-20122 Milano (IT).

(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published:
— *With international search report.*
— *Before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments.*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

WO 01/07014 A1

(54) Title: FORMULATIONS OF STEROID SOLUTIONS FOR INHALATORY ADMINISTRATION

(57) Abstract: The present invention relates to optimized formulations of antiinflammatory steroids for nebulisation and a process for the preparation thereof. More particularly, the invention relates to formulations for monodose or multidose vials in the form of preservative-free stable solutions of a more acceptable osmolarity, which can effectively be nebulised with the nebulisers currently available on the market and are well-tolerated by patients.

10021101 .050297 #6

budesonide

Declaration, Power Of Attorney and Petition

WE (I) the undersigned inventor(s), hereby declare(s) that:

My residence, post office address and citizenship are as stated below next to my name,

We (I) believe that we are (I am) the original, first, and joint (sole) inventor(s) of the subject matter which is claimed and for which a patent is sought on the invention entitled

Formulations of steroid solutions for inhalatory administration

the specification of which

☐ is attached hereto.

☐ was filed on _____ as

Application Serial No. _____

and amended on _____.

☒ was filed as PCT international application

Number PCT/EP00/06916

on 20.07.2000,

and was amended under PCT Article 19

on _____ (if applicable).

We (I) hereby state that we (I) have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

We (I) acknowledge the duty to disclose information known to be material to the patentability of this application as defined in Section 1.56 of Title 37 Code of Federal Regulations.

We (I) hereby claim foreign priority benefits under 35 U.S.C. § 119(a)-(d) or § 365(b) of any foreign application(s) for patent or inventor's certificate, or § 365(a) of any PCT International application which designated at least one country other than the United States, listed below and have also identified below, by checking the box, any foreign application for patent or inventor's certificate, or PCT International application having a filing date before that of the application on which priority is claimed. Prior Foreign Application(s)

Application No.	Country	Day/Month/Year	Priority Claimed
MI99A001625	Italy	23.07.1999	☑ Yes ☐ No
			☐ Yes ☐ No
			☐ Yes ☐ No
			☐ Yes ☐ No

We (I) hereby claim the benefit under Title 35, United States Code, § 119(e) of any United States provisional application(s) listed below.

(Application Number)	(Filing Date)
(Application Number)	(Filing Date)

We (I) hereby claim the benefit under 35 U.S.C. § 120 of any United States application(s), or § 365(c) of any PCT International application designating the United States, listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States or PCT International application in the manner provided by the first paragraph of 35 U.S.C. § 112, I acknowledge the duty to disclose information which is material to patentability as defined in 37 CFR § 1.56 which became available between the filing date of the prior application and the national or PCT International filing date of this application.

Application Serial No.	Filing Date	Status (pending, patented, abandoned)

30

And we (I) hereby appoint: Norman F. Oblon, Reg. No. 24,618; Marvin J. Spivak, Reg. No. 24,913; C. Irvin McClelland, Reg. No. 21,124; Gregory J. Maier, Reg. No. 25,599; Arthur I. Neustadt, Reg. No. 24,854; Richard D. Kelly, Reg. No. 27,757; James D. Hamilton, Reg. No. 28,421; Eckhard H. Kuesters, Reg. No. 28,870; Robert T. Pous, Reg. No. 29,099; Charles L. Gholz, Reg. No. 26,395; Vincent J. Sunderdick, Reg. No. 29,004; William E. Beaumont, Reg. No. 30,996; Robert F. Gnuse, Reg. No. 27,295; Jean-Paul Lavalleye, Reg. No. 31,451; Stephen G. Baxter, Reg. No. 32,884; Robert W. Hahl, Reg. No. 33,893; Richard L. Treanor, Reg. No. 36,379; Steven P. Weihrouch, Reg. No. 32,829; John T. Goolkasian, Reg. No. 26,142; Richard L. Chinn, Reg. No. 34,305; Steven E. Lipman, Reg. No. 30,011; Carl E. Schlier, Reg. No. 34,426; James J. Kulbaski, Reg. No. 34,648; Richard A. Neifeld, Reg. No. 35,299; J. Derek Mason, Reg. No. 35,270; Surinder Sachar, Reg. No. 34,423; Christina M. Gadiano, Reg. No. 37,628; Jeffrey B. McIntyre, Reg. No. 36,867; Paul E. Rauch, Reg. No. 38,591; William T. Enos, Reg. No. 33,128; and Michael E. McCabe, Jr., Reg. No. 37,182; our (my) attorneys, with full powers of substitution and revocation, to prosecute this application and to transact all business in the Patent Office connected therewith; and we (I) hereby request that all correspondence regarding this application be sent to the firm of OBLON, SPIVAK, McCLELLAND, MAIER & NEUSTADT, P.C., whose Post Office Address is: Fourth Floor, 1755 Jefferson Davis Highway, Arlington, Virginia 22202.

We (I) declare that all statements made herein of our (my) own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

1-00

MALVOLTI Chiara
NAME OF FIRST SOLE INVENTOR

Residence: Via Palermo, 26/A
PARMA, Italy IT

Chiara Malvolti
Signature of Inventor

Citizen of: Italy

Post Office Address: same as above

23.01.2002
Date

2-00

GARZIA Raffaella
NAME OF SECOND JOINT INVENTOR

Residence: Via Palermo, 26/A
PARMA, Italy ITX

Signature of Inventor

Citizen of: Italy

Post Office Address: same as above

23.01.2002
Date

3-00

BRAMBILLA Gaetano
NAME OF THIRD JOINT INVENTOR

Residence: Via Palermo, 26/A
PARMA, Italy ITX

Signature of Inventor

Citizen of: Italy

Post Office Address: same as above

23.01.2002
Date

4-00

CHIESI Paolo
NAME OF FOURTH JOINT INVENTOR

Residence: Via Palermo, 26/A
PARMA, Italy ITX.

Signature of Inventor

Citizen of: Italy

Post Office Address: same as above

23.01.2002
Date

NAME OF FIFTH JOINT INVENTOR

Residence:

Signature of Inventor

Citizen of:

Post Office Address:

Date